Institute Report No. 423

Acute Oral Toxicity of 3-Chloro-4,4-dimethyl-2-oxazolidinone (Compound 1) in ICR Mice

Gerald F.S. Hiatt, PhD
Carolyn M. Lewis, MS
Conrad R. Wheeler, PhD
and
Don W. Korte, Jr., PhD, LTC, MSC

MAMMALIAN TOXICOLOGY BRANCH
DIVISION OF TOXICOLOGY

October 1990

Toxicology Series: 164

LETTERMAN ARMY INSTITUTE OF RESEARCH
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129

This document has been approved for public release and sale; its distribution is unlimited.

Destroy this report when it is no longer needed. Do not return to the originator.

Citation of trade names in this report does not constitute an official endorsement or approval of the use of such items.

This research was conducted in compliance with the "Guide for the Care and Use of Laboratory Animals," NIH Publication No. 85-23, as prepared by the Institute of Laboratory Animal Resources, National Research Council.

This material has been reviewed by Letterman Army Institute of Research and there is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the author(s) and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense. *(AR 360-5)*

Donald G. Corby 3 Jan 91

Donald G. Corby **(date)**
COL, MC
Commanding

Unclassified

SECURITY CLASSIFICATION OF THIS PAGE

REPORT DOCUMENTATION PAGE

Form Approved OMB No. 0704-0188

1a. REPORT SECURITY CLASSIFICATION UNCLASSIFIED	1b. RESTRICTIVE MARKINGS
2a. SECURITY CLASSIFICATION AUTHORITY	3 DISTRIBUTION / AVAILABILITY OF REPORT APPROVED FOR PUBLIC RELEASE; DISTRIBUTION IS UNLIMITED.
2b. DECLASSIFICATION / DOWNGRADING SCHEDULE	
4. PERFORMING ORGANIZATION REPORT NUMBER(S) Institute Report No.: 423	5. MONITORING ORGANIZATION REPORT NUMBER(S)
6a. NAME OF PERFORMING ORGANIZATION Mammalian Toxicology Division of Toxicology 6b. OFFICE SYMBOL (If applicable) SGRD-ULE-T	7a. NAME OF MONITORING ORGANIZATION US Army Biomedical Research and Development Laboratory
6c. ADDRESS (City, State, and ZIP Code) Letterman Army Institute of Research Presidio of San Francisco, CA 94129-6800	7b. ADDRESS (City, State, and ZIP Code) Fort Detrick Frederick, MD 21702-5010
8a. NAME OF FUNDING / SPONSORING ORGANIZATION US Army Medical Research & Development Command 8b. OFFICE SYMBOL (If applicable)	9. PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER Project Order 85PP5819/DCRN A150
8c. ADDRESS (City, State, and ZIP Code) Fort Detrick Frederick, MD 21701-5012	10. SOURCE OF FUNDING NUMBERS PROGRAM ELEMENT NO. / PROJECT NO. / TASK NO. / WORK UNIT ACCESSION NO.

11. TITLE (Include Security Classification)
(U) Acute Oral Toxicity of 3-Chloro-4,4-dimethyl-2-oxazolidinone (Compound 1) in ICR Mice

12. PERSONAL AUTHOR(S)
GFS Hiatt, CM Lewis, CR Wheeler, and DW Korte, Jr.

13a. TYPE OF REPORT	13b. TIME COVERED	14. DATE OF REPORT (Year, Month, Day)	15. PAGE COUNT
Institute	FROM JUN85 TO 3JUL85	October 1990	58

16. SUPPLEMENTARY NOTATION
Toxicology Series No. 164

17. COSATI CODES			18. SUBJECT TERMS (Continue on reverse if necessary and identify by block number)
FIELD	GROUP	SUB-GROUP	Acute Oral Toxicity, N-Chloramine, Mouse, Mammalian Toxicology, Water Disinfectant, 3-Chloro-4,4-dimethyl-2-oxazolodinone

19. ABSTRACT (Continue on reverse if necessary and identify by block number)

The acute oral toxicity of 3-chloro-4,4-dimethyl-2-oxazolidinone (Compound 1) was determined in male and female ICR mice by using the oral gavage single-dose method. The median lethal dose was 309 ± 12 mg/kg for male and 332 ± 13 mg/kg for female mice. The predominate clinical observation was a syndrome of generalized malaise which was characterized by inactivity, hunched posture, and rough coat. This syndrome was observed at all dose levels with frequency and severity increasing with dose. One finding of note at the higher dose levels was the presence of hematuria; however, there was no obvious evidence of renal pathology observed at necropsy. Lethality was observed primarily during the first 48 hours after dosing and most of the clinical signs were also observed during this time period. These results place 3-chloro-4,4-dimethyl-2-oxazolidinone in the very toxic category.

20. DISTRIBUTION / AVAILABILITY OF ABSTRACT	21 ABSTRACT SECURITY CLASSIFICATION
☒ UNCLASSIFIED/UNLIMITED ☐ SAME AS RPT ☐ DTIC USERS	UNCLASSIFIED
22a. NAME OF RESPONSIBLE INDIVIDUAL DONALD G. CORBY, COL, MC	22b TELEPHONE (Include Area Code) (415) 561-3600
22c. OFFICE SYMBOL SGRD-ULZ	

DD Form 1473, JUN 86 Previous editions are obsolete SECURITY CLASSIFICATION OF THIS PAGE

Unclassified

ABSTRACT

The acute oral toxicity of 3-chloro-4,4-dimethyl-2-oxazolidinone (Compound 1) was determined in male and female ICR mice by using the oral gavage single-dose method. The median lethal dose was 309 ± 12 mg/kg for male and 332 ± 13 mg/kg for female mice. The predominate clinical observation was a syndrome of generalized malaise which was characterized by inactivity, hunched posture, and rough coat. This syndrome was observed at all dose levels with frequency and severity increasing with dose. One finding of note at the higher dose levels was the presence of hematuria; however, there was no obvious evidence of renal pathology observed at necropsy. Lethality was observed primarily during the first 48 hours after dosing and most of the clinical signs were also observed during this time period. These results place 3-chloro-4,4-dimethyl-2-oxazolidinone in the very toxic category.

KEY WORDS: Acute Oral Toxicity, 3-chloro-4,4-dimethyl-2-oxazolidinone, Mouse, Mammalian Toxicology, Compound 1, Water Disinfectant

PREFACE

TYPE REPORT: Acute Oral Toxicity GLP Study Report

TESTING FACILITY:

US Army Medical Research and Development Command
Letterman Army Institute of Research
Presidio of San Francisco, CA 94129-6800

SPONSOR:

US Army Medical Research and Development Command
US Army Biomedical Research and Development Laboratory
Fort Detrick, MD 21702-5010
Project Officer: James C. Eaton, P.E.

PROJECT ORDER/APC: 85PP5819 Toxicity Studies of Water Disinfectant, Compound 1 (DCRN A150)/TL13

GLP STUDY NUMBER: 85028

STUDY DIRECTOR: LTC Don W. Korte, Jr., PhD, MSC
Diplomate, American Board of Toxicology

PRINCIPAL INVESTIGATOR: Gerald F.S. Hiatt, PhD

CO-PRINCIPAL INVESTIGATOR: John R.G. Ryabik, SP4, BS

PATHOLOGIST: MAJ Michael V. Slayter, DVM, VC

DATA MANAGER: Yvonne C. LeTellier

REPORT/DATA MANAGEMENT: A copy of the final report, study protocol, retired SOPs, raw data, analytical, stability, and purity data of the test compound, tissues, and the test compound will be retained in the LAIR Archives.

TEST SUBSTANCE: 3-Chloro-4,4-dimethyl-2-oxazolidinone

INCLUSIVE STUDY DATES: 5 June 1985 - 3 July 1985

OBJECTIVE: The objective of this study was to determine the acute oral toxicity of 3-chloro-4,4-dimethyl-2-oxazolidinone (Compound 1) in male and female ICR mice.

ACKNOWLEDGMENTS

SSG James D. Justus, BS, SP4 James J. Fischer, and SP4 Scott L. Schwebe provided research assistance and animal care and Colleen S. Kamiyama and Ann L. Wilkinson provided secretarial assistance.

SIGNATURES OF PRINCIPAL SCIENTISTS AND MANAGERS INVOLVED IN THE STUDY

We, the undersigned, declare that GLP Study 85028 was performed under our supervision, according to the procedures described herein, and that this report is an accurate record of the results obtained.

Don W. Korte Jr. 23Oct90
DON W. KORTE JR., PhD/DATE
LTC, MS
Study Director

Don W. Korte Jr. 23Oct90
GERALD F.S. HIATT, PhD/DATE
DAC
Principal Investigator

Don W. Korte Jr. 23Oct90
JOHN R.G. RYABIK, BS/DATE
SP4, USA
Co-Principal Investigator

Conrad Wheeler 30 Oct 90
CONRAD R. WHEELER, PhD/DATE
DAC
Chemist

Yvonne C LeTellier 30 Oct 90
YVONNE C. LETELLIER, BS/DATE
DAC
Data Manager

Carolyn M. Lewis 5 Dec 90
CAROLYN M. LEWIS, MS/DATE
DAC
Technical Report Writer

DEPARTMENT OF THE ARMY
LETTERMAN ARMY INSTITUTE OF RESEARCH
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129-6800

REPLY TO
ATTENTION OF:

SGRD-ULX-QA

29 October 1990

MEMORANDUM FOR Commander, LAIR

SUBJECT: QAU Clearance of Report for GLP Study 85028
Tox Series 164

I have reviewed the reply to the QAU audit conducted on 4 October 1990 and find it to be acceptable. I believe the report accurately reflects the raw data and recommend it be approved for publication.

Walter G. Bell

WALTER G. BELL
SFC, USA
Quality Assurance Officer

TABLE OF CONTENTS

Page

Abstract.....i

Preface.....iii

Acknowledgments.....iv

Signatures of Principal Scientists.....v

Report of Quality Assurance Unit.....vi

Table of Contents.....vii

INTRODUCTION.....1

Objective of Study.....1

MATERIALS.....1

Test Substance.....1
Vehicle.....2
Animal Data.....2
Husbandry.....2

METHODS.....2

Group Assignment/Acclimation.....2
Dose Levels.....3
Compound Preparation.....3
Chemical Analyses.....3
Test Procedures.....4
Observations.....4
Necropsy.....4
Statistical Analyses.....4
Duration of Study.....5
Changes/Deviations.....5
Raw Data and Final Report Storage.....5

RESULTS.....5

Mortality.....5
Lethal Dose Calculation 5
Clinical Observations.....8
Gross Pathology Observations.....12

TABLE OF CONTENTS (cont.)

DISCUSSION.....12

CONCLUSION.....12

REFERENCES.....13

APPENDICES.....14

Appendix A. Chemical Data.....15
Appendix B. Animal Data.....20
Appendix C. Historical Listing of Study Events.....21
Appendix D. Cumulative Mortality Data.....22
Appendix E. Individual Animal Histories.....23
Appendix F. Individual Body Weights.....40
Appendix G. Pathology Report.....52

OFFICIAL DISTRIBUTION LIST.....58

INTRODUCTION

The U.S. Air Force has sponsored development of a new disinfectant for field water supplies. HTH (high-test hypochlorite), the existing field water disinfectant, is a mixture of calcium hypochlorite and inert ingredients; however, it is not stable under field or storage conditions. In addition, HTH produces carcinogenic trihalomethanes upon reaction with organic impurities in the water. N-chloramines are being evaluated as water disinfectants because of their wide-spectrum disinfectant qualities, exceptional stability, and minimal potential to produce trihalomethanes (1). The U.S. Army Biomedical Research and Development Laboratory (USABRDL) has been assigned the mission of evaluating the "health effects" of a promising N-chloramine, 3-chloro-4,4-dimethyl-2-oxazolidinone (Compound 1). As part of their mandate, USABRDL has tasked the Division of Toxicology, Letterman Army Institute of Research (LAIR), to develop an acute toxicity profile for Compound 1. Evaluation of its acute oral toxicity is an integral part of the acute toxicity profile for Compound 1.

Objective of Study

The objective of this study was to determine the acute oral toxicity of Compound 1 in male and female ICR mice.

MATERIALS

Test Substance

Chemical Name: 3-Chloro-4,4-dimethyl-2-oxazolidinone

Chemical Abstracts Service Registry No.: 58629-01-9

LAIR Code Number: TP55

Molecular Formula: $C_5H_8ClNO_2$

Other test substance information is presented in Appendix A.

Vehicle

The vehicle for 3-chloro-4,4-dimethyl-2-oxazolidinone was 1% gum tragacanth (Lot No. 34F-0156, Sigma Chemical Company, St. Louis, MO) in sterile water for injection (Lot No. 62-354-DM-03, Abbott Labs, North Chicago, IL). The expiration date was 1 March 1986.

Animal Data

Eighty male and 80 female ICR mice (Harlan Sprague-Dawley, Inc., Indianapolis, IN) were used for this study. They were identified individually with tags numbered 85C00514 - 85C00673 (inclusive) placed in the dorsal skin of the neck. Two males and 2 females were selected at random for quality control necropsy evaluation at receipt. Thirty-six of the animals were used in an Approximate Lethal Dose (ALD) determination and two were not dosed. The animal weights on receipt (6 Jun 85) ranged from 20 to 36 g. Additional animal data appear in Appendix B.

Husbandry

Mice were caged individually in stainless steel wire mesh cages in racks equipped with automatic flushing dumptanks. The diet, fed *ad libitum*, consisted of Certified Purina Rodent Chow Diet 5002 (Ralston Purina Company, St. Louis, MO); water was provided by lixit valves on a central line. The animal room temperature was constantly monitored and maintained in a range from 21.7°C to 23.3°C with a relative humidity range of 41% to 48%. The photoperiod was 12 hours of light per day.

METHODS

Group Assignment/Acclimation

Study mice were randomized into five dose groups and a vehicle control group of 10 males and 10 females each. Allocation was accomplished by using a computer-based stratified weight-biased method. The Beckman TOXSYS® Animal Allocation Program was used in conjunction with a Beckman TOXSYS® Data Collection Terminal. The animals were acclimated for 15-17 days before the day of dosing. During this period they were observed daily for signs of illness.

Dose Levels

The results of the ALD determination suggested that the median lethal dose (MLD) was below 250 mg/kg for the males and near 250 mg/kg for the females. Based on these data, test doses were selected (Table 1).

TABLE 1: Group Dose Schedule

Group	Dose (mg/kg)	
	Females	Males
1	159	200
2	251	316
3	398	501
4	316	251
5	501	398
6	vehicle	vehicle

Compound Preparation

Compound 1 was received in a solid form. It was ground in a freezer mill (Model 6700, Spex Industries, Inc., Metuchen, NJ). Compound 1 was then suspended in 1% gum tragacanth using a Brinkman Polytron Homogenizer (Brinkman Instruments Co., Westbury, NY) for mixing, which resulted in a milky, viscous liquid.

Chemical Analyses

The identity of Compound 1 was verified by comparison of IR spectra obtained on receipt with the spectra provided by the source (Appendix A). The purity of the test substance was determined by analyzing four pure samples by iodometric titration. The purity was approximately 99%. Analysis of an aqueous suspension of Compound 1 (50 mg/ml) in 1% gum tragacanth by iodometric titration demonstrated that it was stable in water for at least three days. Formulating Compound 1 in 1% gum tragacanth produced a homogenous suspension over a range of 18 to 160 mg/ml. Accuracy of the Compound 1 suspensions ranged from 94.9% to 106.9% of target.

Test Procedures

This study was conducted in accordance with EPA guidelines (2) and LAIR SOP OP-STX-36 (3). The volume of dosing solution given each animal was based upon the desired dose level and the compound concentration in suspension. The dose level was increased by varying the concentration of each suspension, and animals received calculated volumes based upon weight. Volumes ranged from 0.27 to 0.43 ml in the male and 0.22 to 0.36 ml in female mice. The vehicle control group was given 0.25 to 0.35 ml of the vehicle. Dosing was performed by oral gavage without animal sedation or anesthesia. Sterile, disposable syringes (Becton, Dickenson & Co., Rutherford, NJ) fitted with 20-gauge, 1-1/2-inch, ball-tipped, stainless steel Perfektum® oral gavage animal tubes (Popper & Sons, Inc., New Hyde Park, NY) were used for dosing. Animals in Groups 1 - 3 were dosed on 17 June 1985. Animals in Groups 4 - 6 were dosed on 19 June 1985 after analysis of the data from 17 June.

Observations

Observations for mortality and signs of acute toxicity were performed daily according to the following procedure: 1) animals were observed undisturbed in their cages, 2) animals were removed from their cages and given a physical examination, and 3) animals were observed after being returned to their cages. On the day of dosing, the mice were checked intermittently throughout the day. Recorded observations were performed approximately 2, and 4 hours after dosing, and daily for the remainder of the 2-week test period. A second "walk-through" observation was performed daily with only significant observations recorded. Body weights were recorded periodically during the course of the study.

Necropsy

Animals that died during the observation period were submitted for necropsy. Those that survived the 14-day study period were also submitted for necropsy after termination by barbiturate overdose.

Statistical Analyses

Statistical analyses were performed on the study results. Selected lethal doses for the females were derived by probit analysis using the maximum likelihood method, as described by Finney (4) and for the males the median lethal dose was calculated manually by the moving average method

(5). The program, PROBIT, developed for the Data General Computer, Model MV8000, was used to draw the probit curve and lethal dose values.

Duration of Study

Appendix C is a historical listing of study events.

Changes/Deviations

The vehicle was changed from sterile water to 1% gum tragacanth (in sterile water) because Compound 1 was not completely soluble in water at concentrations necessary for dosing. The change in the vehicle required increasing the number of animals assigned to the vehicle control group because 1% gum tragacanth had not previously been used in this laboratory for a mouse study. Probit analysis was not used to calculate the MLD in male mice because there was only one dose level with a fractional response. The moving average method was used instead and provided an MLD determination. The additional information to be obtained from the probit analysis was not deemed sufficient to warrant the use of additional animals in the study.

Raw Data and Final Report Storage

A copy of the final report, study protocols, raw data, all SOPs, and an aliquot of the test compound will be retained in the LAIR Archives.

RESULTS

Mortality

Forty-nine animals died as a result of dosing with Compound 1. Twenty-nine deaths occurred within 24 hours after dosing. An additional 12 deaths occurred between 24 and 48 hours after dosing. Table 2 lists the compound-related deaths by group. Appendix D is a tabular presentation of cumulative mortality.

Lethal Dose Calculation

Lethal dose values were calculated by probit analysis for the female mice. The equation for the probit regression line was $Y = -40.16 + 17.92 \log X$, where X is the dose and Y the corresponding probit value. Lethal doses calculated from the equation for the probit regression line are presented in Table 3. Figure 1 graphically presents the regression line for the female mice.

TABLE 2: Compound-Related Deaths by Group

Group	Dose (mg/kg)	Compound-Related Death/ Number in Group	Percent Mortality
		MALES	
1	200	0/9*	0
2	251	0/10	0
4	316	6/10	60
3	398	10/10	100
5	501	10/10	100
6	Vehicle	0/10	0
		FEMALES	
1	159	0/9*	0
4	251	0/10	0
2	316	4/10	40
5	398	9/10	90
3	501	10/10	100
6	Vehicle	0/10	0

* One animal was not used because it was not fasted.

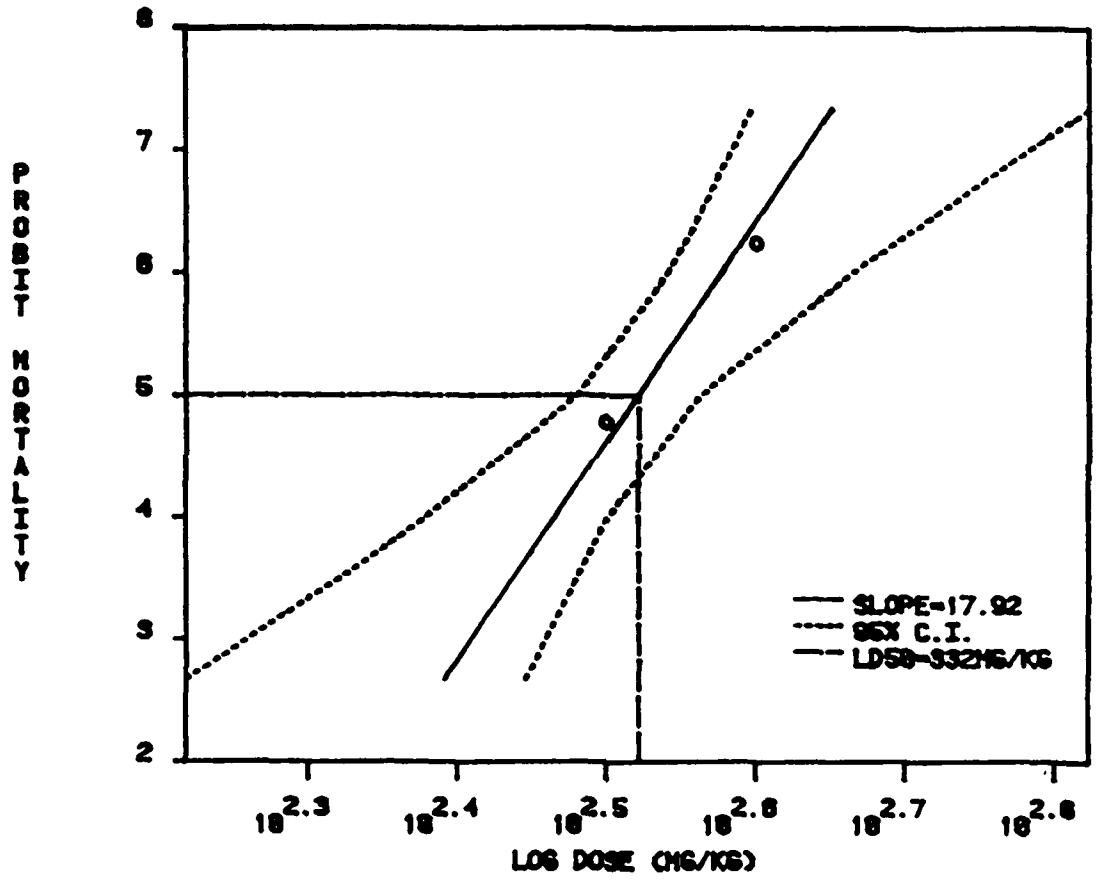
TABLE 3: Calculated Lethal Doses (LD) of Compound 1 in ICR Mice

Effect Level	Calculated Dose* (mg(base)/kg)	95% Confidence Limits (mg(base)/kg)
	MALES	
LD50	309 ± 12	(289, 333)
	FEMALES	
LD10	281 ± 17	(222, 308)
LD50	332 ± 13	(301, 365)
LD90	391 ± 24	(358, 497)

*Lethal Dose ± standard error.

Figure 1

Dose Response Curve for 3-Chloro-4,4-dimethyl-2-oxazolidinone in Female Mice



Since probit analysis requires a fractional response in more than one group, the MLD and 95% confidence limits for the male mice were calculated by moving averages. This method involves interpolation and does not estimate a regression line. Consequently, the other lethal dose values and their confidence limits could not be calculated.

Clinical Observations

A variety of clinical signs were observed following oral administration of Compound 1. The predominate clinical signs were in the behavioral category (inactivity, tremors, jumping, and hypotonia) which was observed in 76 of 98 animals dosed and hunched posture which was observed in 64 of 98 animals. Urogenital signs (bloody urine, red material on the abdomen, and bloody penis) were observed in 28 of 98. Sixteen animals were cool to the touch (decreased temperature). Of the 49 males dosed with Compound 1, 30 exhibited a rough coat and 17 exhibited yellow stains of the abdomen and perianal area. Generally, the incidence, severity, and duration of signs exhibited a dose-response relationship, although the early deaths of animals in the high dose group masked this relationship.

Other clinical signs included diarrhea, depressed grasping and righting reflex, gasping, increased respiratory rate, lacrimation, and prostration and were observed in varying combinations in nine animals.

The incidence of clinical signs was greatest during the first two observations following dosing. Most of the surviving animals were still exhibiting some signs 24 hours after dosing. Only a few animals continued to exhibit signs after the first two days. The signs that were observed in these few animals included hunched posture, inactivity, rough coat, and yellow stains on the abdomen and perianal area. All of these signs except rough coat disappeared by 7 days after dosing. Tables 4 and 5 contain summaries of the clinical signs observed in the male and female mice, respectively. Appendix E contains the individual animal histories.

Weight gains of survivors were not affected by dosing. Table 6 presents the mean body weights by groups. Appendix F contains individual weight tables.

TABLE 4: Incidence Summary for Clinical Observations in Male Mice Administered Compound 1

Category of Clinical Signs	Group / Dose (mg/kg) / N=	6 Control 10	1 200 9	4 251 10	2 316 10	5 398 10	3 501 10
Behavioral[a]		0	7	10	10	10	10
Hunched Posture		1	8	4	10	2	10
Rough Coat		1	5	4	5	9	7
Urogenital[b]		-	-	1	1	8	6
Decreased Temperature		-	-	-	-	6	2
Stains[c]		1	3	4	4	3	3
Other[d]		-	-	-	1	2	1
Death		-	-	-	6	10	10
Normal		8	-	-	-	-	-

[a] Includes inactivity, tremors, jumping, and hypotonia.

[b] Includes bloody urine/feces, bloody penis, and red material on abdomen.

[c] Includes yellow/brown stains or material on mouth, abdomen, or perianal area and stains on the mouth and nose.

[d] Includes diarrhea, depressed grasping and/or righting reflexes, and gasping.

TABLE 5: Incidence Summary for Clinical Observations in Female Mice Administered Compound 1

Category of Clinical Signs	Group / Dose (mg/kg) / N=	6 Control 10	1 159 9	2 251 10	4 316 10	3 398 10	5 501 10
Behavioral[a]		-	2	1	8	10	8
Hunched Posture		-	6	5	5	10	4
Urogenital[b]		-	-	-	4	5	3
Decreased Temperature		-	-	-	2	5	1
Stains[c]		-	1	-	1	2	-
Other[d]		-	-	-	2	1	2
Death		-	-	-	4	9	10
Death (no signs recorded)		-	-	-	-	-	2
Normal		10	2	5	-	-	-

[a] Includes inactivity, tremors, jumping, and hypotonia.

[b] Includes bloody (red) urine/feces and red material on abdomen.

[c] Includes yellow stains/material on mouth and red stains on the nose.

[d] Includes diarrhea, depressed grasping and/or righting reflexes, gasping, increased respiratory rate, lacrimation, and prostration.

TABLE 6: Mean Body Weights in Grams ± S.E †

Dose (mg/kg)	Receipt	Dosing	Day 7	Day 14
		MALES		
200	28.4 ±1.1(9)	32.4 ±0.9(9)	35.7 ±0.9(9)	36.2 ±0.7(9)
251	29.4 ±1.1(10)	33.4 ±0.8(10)	33.6 ±0.5(10)	35.3 ±0.4(10)
316	29.2 ±1.2(10)	33.1 ±0.8(10)	34.2 ±1.3(4)	36.7 ±0.9(4)
398	29.3 ±0.7(10)	34.1 ±1.0(10)	---	---
501	29.3 ±0.9(10)	33.4 ±0.8(10)	---	---
Vehicle	29.4 ±0.9(10)	32.7 ±0.7(10)	36.3 ±0.9(10)	36.0 ±0.6(10)
		FEMALES		
159	27.8 ±1.0(9)	29.2 ±0.8(9)	30.6 ±0.5(9)	30.9 ±0.7(9)
251	26.5 ±1.0(10)	28.6 ±1.0(10)	29.4 ±0.7(10)	29.7 ±0.8(10)
316	25.2 ±0.6(10)	27.4 ±0.4(10)	28.8 ±0.7(6)	27.8 ±0.9(6)
398	27.7 ±0.7(10)	29.1 ±0.8(10)	24.0(1)	27.0(1)
501	26.8 ±0.5(10)	28.5 ±0.7(10)	---	---
Vehicle	28.2 ±0.9(10)	29.6 ±0.8(10)	30.0 ±1.0(10)	31.0 ±0.9(10)

† Number in parentheses = number of animals.

Gross Pathology Observations

Among the 49 animals that died after administration of Compound 1, none had any remarkable lesions that could be attributed to the test compound. The veterinary pathologist's report appears at Appendix G.

DISCUSSION

The calculated MLD for Compound 1 was 309 ± 12 mg/kg in male and 332 ± 13 mg/kg in female ICR mice. These MLD values place Compound 1 in the very toxic range (6).

Compound 1, when administered by oral intubation, produced a syndrome that could be described as a failure to thrive and/or generalized malaise as approximately 75% of the treated animals were inactive and exhibited hunched posture and, in males, rough coat. These signs were observed in all dose groups with increasing frequency and severity with dose. As the dose increased, other signs were also observed. These included tremors, hematuria (bloody urine, red material on abdomen, and bloody penis), and decreased temperature. The tremors and decreased temperature could be attributed to the marked inactivity observed in the animals that received the higher doses. The hematuria suggests an effect on the renal system; however, there was no evidence observed at necropsy which would confirm or disprove this hypothesis.

Although these data suggest that Compound 1 is a very toxic compound, it also indicates that the toxicity should have little effect on the proposed use of Compound 1 as a water disinfectant. As a water disinfectant, Compound 1 is effective at a concentration of 210 parts per million against the most resistant bacteria (7), a concentration several orders of magnitude lower than the concentrations used in this study.

CONCLUSION

3-Chloro-4,4-dimethyl-2-oxazolidinone (Compound 1) is a very toxic compound which produces a nonspecific syndrome of malaise and a failure to thrive. Calculated median lethal dose values were 309 mg/kg in male and 332 mg/kg in female ICR mice.

REFERENCES

1. Worley SD, Swango LJ, Attleberger MH, *et al.* New disinfection agents for water. Tyndall Air Force Base, Florida: Air Force Engineering and Services Center, 1984, Engineering and Services Laboratory Publication No. ESL-TR-83-68, p 1.

2. Environmental Protection Agency. Office of Pesticides and Toxic Substances, Office of Toxic Substances (TS-792). Acute exposure, oral toxicity. In: Health effects test guidelines, Washington, DC: Environmental Protection Agency, August 1982; EPA 560/6-82-001.

3. Acute oral toxicity study (ALD and LD_{50}). LAIR Standard Operating Procedure OP-STX-36, Letterman Army Institute of Research, Presidio of San Francisco, CA. 15 June 1984.

4. Finney DJ. Probit analysis. 3rd ed. Cambridge: Cambridge University Press, 1971:20-80.

5. Thompson WR. Use of moving averages and interpolation to estimate median-effective dose. Bacteriol. Rev. 1947; 11:115-145.

6. Hodge HC, Sterner JH. Tabulation of toxicity classes. Amer Ind Hyg Assoc Q 1943; 10:93-96.

7. Mora EC, Kohl HH, Wheatley WB, *et al.* Properties of a new chloramine disinfectant and detoxicant. Poultry Sci 1982; 61:1968-1971.

Page

Appendix A. Chemical Data.....15

Appendix B. Animal Data.....20

Appendix C. Historical Listing of Study Events.....21

Appendix D. Cumulative Mortality Data.....22

Appendix E. Individual Animal Histories.....23

Appendix F. Individual Body Weights.....40

Appendix G. Pathology Report.....52

Appendix A: CHEMICAL DATA

Chemical Name: 3-Chloro-4,4-dimethyl-2-oxazolidinone

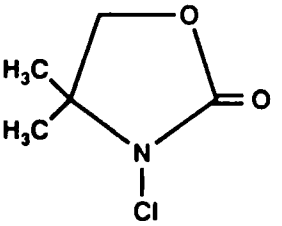
Other Names: Compound 1, Agent 1

Lot Number: 8

Chemical Abstracts Service Registry No.: 58629-01-9

LAIR Code No.: TP55

Structural Formula:



Molecular Formula: $C_5H_8ClNO_2$

Molecular Weight: 149.5

Physical State: White crystalline solid

Purity: 98.95%

Melting point: 68-70°C

IR: (KBR) 3499, 2973, 2932, 2877, 1760, 1512, 1457, 1395, 1371, 1291, 1265, 1215, 1176, 1056, 1017, 999, 958, 938, 843, 757, 693 cm^{-1}. The IR spectrum obtained on receipt of the compound was identical to that provided by the source.[1]

Stability: An aqueous suspension of Compound 1 (50 mg/ml) at room temperature is stable for at least 3 days.[2]

Source: Dr. S.D. Worley
Dept. of Chemistry
Auburn University
Auburn, Alabama

---

[1] Wheeler CR. Nitrocellulose-Nitroguanidine Projects. Laboratory Notebook #84-05-101.3, p. 26. Letterman Army Institute of Research, Presidio of San Francisco, CA.
[2] Ibid. p. 42.

Appendix A (cont.): CHEMICAL DATA

ANALYSIS OF DOSING SUSPENSIONS

Aqueous suspensions of Compound 1 in 1% gum tragacanth were prepared by mixing with a Brinkman Polytron homogenizer. The suspensions were subsequently used for GLP studies 85027 (acute oral toxicity in rats), 85028 (acute oral toxicity in mice), and 85032 (dermal sensitization in guinea pigs). After dosing the suspensions were stored at 4°C until analyzed. The concentration of Compound 1 was determined by iodometric titration.

Materials

The following chemicals were obtained from the sources specified:

1. Potassium Iodide, A.R.
   Mallinckrodt Chemical Works, St. Louis, MO
   Lot No. SHK

2. Sodium Thiosulfate, Anhydrous
   J.T. Baker Chemical Co., Phillipsburg, NJ
   Lot No. 5363020

3. Starch, Soluble, Certified A.C.S.
   Fisher Scientific, Fairlawn, NJ
   Lot No. 786070

4. Potassium Iodate, A.C.S.
   Fisher Scientific, Fairlawn, NJ
   Lot No. 790539

5. Sodium Carbonate, Anhydrous
   J.T. Baker Chemical Co., Phillipsburg, NJ
   Lot No. 6161629

Methods

The iodometric titration was performed according to the method described by Skoog and West[1], except for the addition of ethanol to facilitate the dissolution of Compound 1 in water before titration. Using this method, excess iodide is added to a solution of Compound 1. The iodide ion

---

[1] Skoog DA, West DM. Fundamentals of Analytical Chemistry, 4th Ed. Philadelphia: Saunders College Publishing, 1982: 386-388, 764-767.

Appendix A (cont.): CHEMICAL DATA

ANALYSIS OF DOSING SUSPENSIONS (cont.)

Methods (cont.)

(moderately strong reducing agent) reacts with Compound 1 (oxidant), producing iodine or iodine monochloride, which then is titrated with sodium thiosulfate (oxidant). The solution changes from brown to pale yellow as the end point is reached. Before titrating the samples, the molarity of the thiosulfate solution is determined (i.e., standardized) using potassium iodate. Since two thiosulfate ions react with one iodine molecule, the number of moles of Compound 1 in the solution can be determined from the volume of standardized thiosulfate solution added to reach the end point.

Moles Compound 1 = M $Na_2S_2O_3$ / 2 **X** ml $Na_2S_2O_3$ / 1000 ml/l

By entering values for the number of moles of Compound 1 in the solution, the molecular weight of Compound 1 and the volume of sample added in the equation below, the concentration (mg/ml) of the dosing suspensions can be calculated.

Concentration =

moles Compound 1 **X** MW Compound 1 **X** 1000 mg/g / ml Sample

To assess the homogeneity of the dosing suspensions, three 1 ml aliquots were analyzed from the top (T), middle (M), and bottom (B) of three dosing suspensions representing the range of concentrations prepared.

Results

The results from the analysis of the dosing suspensions are presented in Table 1. In Table 2 are the results from the homogeneity determinations.

Discussion

Results from the analysis of the dosing suspensions (Table 1), indicated that two of the suspensions (100 mg/ml and 160 mg/ml) used in GLP Study 85027 deviated by more than 10% from the target values. All other samples were within 6.9% of their target concentrations. The 100 mg/ml and 160 mg/ml

Appendix A (cont.): CHEMICAL DATA

ANALYSIS OF DOSING SUSPENSIONS (cont.)

Discussion (cont.)

suspensions from GLP Study 85027 and the 18 mg/ml suspension from GLP Study 85028 were then analyzed for homogeneity (Table 2). Regardless of the sampling site (top, middle, or bottom of the suspension), the mean deviation from the actual mean of the suspension did not exceed 2.3%. These data indicate that homogeneous suspensions of Compound 1 could be made using 1% gum tragacanth.

Table 1: Analysis of Compound 1 Dosing Suspensions*†

GLP Study Number	Target Conc. (mg/ml)	Date Prepared	Date Analyzed	Actual Conc. (mg/ml)	Percent of Target Conc.
85027	64	8/7/85	13/8/85	62.2	97.2
	100	8/7/85	13/8/85	115.2	115.2
	160	8/7/85	13/8/85	186.4	116.5
85028	18	17/6/85	13/8/85	18.4	102.2
	28	17/6/85	13/8/85	27.4	97.9
	28	19/6/85	13/8/85	29.6	105.7
	45	17/6/85	13/8/85	42.7	94.9
	45	19/6/85	13/8/85	48.1	106.9
85032	10	23/9/85	5/12/85	10.3	103.4
	10	1/10/85	5/12/85	10.3	103.2
	10	7/10/85	5/12/85	10.4	104.1
	10	21/10/85	5/12/85	9.9	99.1

* Wheeler CW. Nitrocellulose-nitroguandine projects. Laboratory Notebook #85-050-010.3, p. 6-16. Letterman Army Institute of Research, Presidio of San Francisco, CA.

† Wheeler CW. Toxicity studies of water disinfectant. Laboratory Notebook #85-12-021, p. 1-8. Letterman Army Institute of Research, Presidio of San Francisco, CA.

Appendix A (cont.): CHEMICAL DATA

ANALYSIS OF DOSING SUSPENSIONS (cont.)

Table 2: Assessment of Homogeneity for Compound 1 Dosing Suspensions*

Target Conc. (mg/ml)	Sampling Site†	Actual Conc. (mg/ml)	Mean Site Conc. (mg/ml)	Mean Suspension Conc. (mg/ml)	Deviation from Suspension Mean (%)
18	T1	18.6			
	T2	18.5	18.4		0.0
	T3	18.1			
	M1	18.3			
	M2	18.1	18.3	18.4	-0.1
	M3	18.5			
	B1	18.6			
	B2	18.6	18.6		0.2
	B3	18.6			
100	T1	114.1			
	T2	114.0	114.3		-0.9
	T3	114.8			
	M1	115.4			
	M2	112.6	114.5	115.2	-0.7
	M3	115.6			
	B1	116.3			
	B2	116.7	116.7		1.5
	B3	117.0			
160	T1	188.4			
	T2	178.7	185.2		-1.2
	T3	188.5			
	M1	186.1			
	M2	187.9	185.5	186.4	-0.9
	M3	182.4			
	B1	188.5			
	B2	190.9	188.7		2.3
	B3	186.7			

* Wheeler CW. Nitrocellulose-nitroguanidine projects. Laboratory Notebook #85-050-010.3, p 6-16. Letterman Army Institute of Research, Presidio of San Francisco, CA.

† Three samples were withdrawn from the top (T), middle (M), and bottom (B) of the suspension.

Appendix B: ANIMAL DATA

Species: *Mus musculus*

Strain: ICR

Source: Harlan Sprague-Dawley, Inc.
P.O. Box 29176
Indianapolis, IN 46229

Sex: Male and female

Date of birth: Males: 26 April 1985
Females: 5 April 1985

Method of randomization: Weight bias, stratified animal randomization using the TOXSYS Animal Allocation Program (SOP OP-ISG-24)

Animals in each group: 10 male and female animals.

Condition of animals at start of study: Normal

Body weight range at dosing: Males: 29 - 41 g
Females: 24 - 36 g

Identification procedures: Cervical tag, tag numbers 85C00514 to 85C00673, inclusive.

Pretest conditioning: Quarantine/acclimation 4-16 June 1985

Justification: The laboratory mouse has proven to be a sensitive and reliable system for lethal dose determination.

Appendix C: HISTORICAL LISTING OF STUDY EVENTS

Date	Event
5 Jun 85	Received 80 male and 80 female ICR mice. Mice were checked for physical condition, sexed, and individually caged.
5-16 Jun 85	Animals were observed daily during quarantine/acclimation period.
6 Jun 85	Animals were tagged and weighed. Four were submitted for necropsy quality control.
13 Jun 85	Animals were weighed and randomized into dose groups.
14 Jun 85	Phase 1 animals were removed from quarantine and weighed.
17 Jun 85	Food was removed from Phase 1 animals cages at 0600 hours. Animals were weighed and dosed at approximately 1100 hours. Observations were recorded approximately 2 and 4 hours after dosing.
18 Jun 85	Phase 2 animals were removed from quarantine and weighed. Phase 1 animals were observed in a.m. and p.m.
19-31 Jun 85	Phase 1 animals were observed in the a.m. and p.m.
19 Jun 85	Food was removed from Phase 2 animals cages at 0600 hours. Animals were dosed at approximately 1100 hours. Observations were recorded approximately 2 and 4 hours after dosing
20 Jun-2 Jul 85	Phase 1 animals were observed in the a.m. and p.m.
24 Jun 85	Phase 1 animals were weighed.
25 Jun 85	Phase 2 animals were weighed.
1 Jul 85	Food was removed from Phase 1 animals cages at 0600 hours. Phase 1 animals were observed, weighed and submitted for necropsy.
3 Jul 85	Food was removed from Phase 2 animals cages at 0600 hours. Phase 2 animals were observed, weighed and submitted for necropsy.

Appendix D: CUMULATIVE MORTALITY DATA*

Dose	Animals Dosed	Time after Dosing: Hours 2	Hours 4	Days 1	2	3	4	5	6	7-14
				MALES						
0	10	0	0	0	0	0	0	0	0	0
200	9	0	0	0	0	0	0	0	0	0
251	10	0	0	0	0	0	0	0	0	0
316	10	0	0	3	4	6	6	6	6	6
398	10	0	0	3	9	9	9	9	10	10
501	10	0	1	10	10	10	10	10	10	10
				FEMALES						
0	10	0	0	0	0	0	0	0	0	0
159	9	0	0	0	0	0	0	0	0	0
251	10	0	0	0	0	0	0	0	0	0
316	10	0	0	1	3	4	4	4	4	4
398	10	0	0	2	5	8	9	9	9	9
501	10	0	1	10	10	10	10	10	10	10
Totals		0	2	29	41	47	48	48	49	49

* Values are deaths/dose group.

Appendix E: INDIVIDUAL ANIMAL HISTORIES

MALE: 200 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00551	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
85C00553	Rough Coat	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
85C00563	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Slight
	Stain, Yellow, Abdomen	Jun 17-20	Slight
85C00567	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
	Stain, Yellow, Mouth	Jun 17	Slight
	Rough Coat	Jun 17	Moderate
85C00573	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Slight
	Jumping	Jun 17	Slight
85C00576	Hunched Posture	Jun 17	Slight
85C00582	Inactive	Jun 17	Slight
85C00587	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
	Stain, Yellow, Mouth	Jun 17	Slight
	Rough Coat	Jun 17	Slight
85C00591	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: 251 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00547	Urine, Bloody	Jun 19	Present
	Feces, Bloody	Jun 19	Present
	Inactive	Jun 19	Slight
85C00556	Inactive	Jun 19	Moderate
	Rough Coat	Jun 19	Slight
	Hunched Posture	Jun 20-26	Slight
85C00565	Inactive	Jun 19	Moderate
	Rough Coat	Jun 19	Slight
	Stain, Yellow, Abdomen	Jun 20	Slight
85C00566	Inactive	Jun 19	Slight
	Jumping	Jun 19	Slight
85C00568	Inactive	Jun 19	Moderate
	Material, Yellow, Perianal	Jun 19,20	Slight
	Stain, Yellow, Perianal	Jun 22,23	Slight
85C00569	Inactive	Jun 19	Moderate
	Rough Coat	Jun 19-25	Moderate
	Material, Yellow, Perianal	Jun 19,20	Moderate
	Stain, Yellow, Perianal	Jun 22,23	Slight
85C00575	Inactive	Jun 19	Slight
	Rough Coat	Jun 19-30	Moderate
	Material, Yellow, Perianal	Jun 19-22	Moderate
	Stain, Yellow, Perianal	Jun 23	Slight
85C00585	Inactive	Jun 19	Slight
	Hunched Posture	Jun 19,20	Slight
85C00592	Inactive	Jun 19	Slight
	Hunched Posture	Jun 19	Slight
85C00593	Inactive	Jun 19	Slight
	Hunched Posture	Jun 19	Slight

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: 316 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00534	Inactive	Jun 17,18	Moderate
	Hunched Posture	Jun 17,18	Slight
	Rough Coat	Jun 17,18	Slight
	Tremors	Jun 18	Slight
	Death	Jun 19	2.1 d
85C00535	Inactive	Jun 17,18	Slight
	Hunched Posture	Jun 17,18	Slight
85C00537	Inactive	Jun 17,18	Moderate
	Rough Coat	Jun 17,29,30	Slight
	Hunched Posture	Jun 17,18	Slight
	Material, Brown, Abdomen	Jun 17	Slight
	Stain, Yellow, Perianal	Jun 22,23	Slight
85C00540	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Material, Red, Abdomen	Jun 17	Slight
	Penis, Bloody	Jun 17	Present
	Death	Jun 18	19.7 h
85C00542	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Slight
	Death	Jun 18	19.7 h
85C00543	Rough Coat	Jun 17	Slight
	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Death	Jun 18	19.7 h
85C00550	Inactive	Jun 17,18	Slight
	Hunched Posture	Jun 17,18	Slight
	Rough Coat	Jun 17	Slight
	Stain, Yellow, Mouth	Jun 17	Slight
	Stain, Yellow, Perianal	Jun 22,23	Slight
85C00560	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17,18	Slight

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: 316 mg/kg Compound 1 (cont.)

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00574	Inactive	Jun 17-19	Marked
	Hunched Posture	Jun 17,18	Slight
	Stain, Yellow, Abdomen	Jun 18	Slight
	Hypotonia	Jun 19	Moderate
	Depr. Righting Reflex	Jun 19	Moderate
	Depr. Grasping Reflex	Jun 19	Moderate
	Death	Jun 20	2.8 d
85C00586	Inactive	Jun 17,18	Moderate
	Hunched Posture	Jun 17	Slight
	Tremors	Jun 17,18	Moderate
	Stain, Yellow, Abdomen	Jun 18	Slight
	Death	Jun 19	1,8 d

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: 398 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00533	Inactive	Jun 19-24	Slight
	Hunched Posture	Jun 19	Slight
	Jumping	Jun 19	Slight
	Rough Coat	Jun 20-24	Slight
	Stain, Yellow, Perianal	Jun 22-24	Slight
	Death	Jun 25	5.9 d
85C00539	Inactive	Jun 19,20	Moderate
	Hypotonia	Jun 19,20	Slight
	Rough Coat	Jun 19	Slight
	Tremors	Jun 20	Moderate
	Material, Red, Abdomen	Jun 20	Moderate
	Urine, Bloody	Jun 20	Present
	Decr. Temperature	Jun 20	N/A
	Death	Jun 20	1.2 d
85C00541	Inactive	Jun 19,20	Moderate
	Hunched Posture	Jun 19,20	Slight
	Rough Coat	Jun 19	Slight
	Tremors	Jun 20	Slight
	Material, Red, Abdomen	Jun 20	Slight
	Decr. Temperature	Jun 20	N/A
	Death	Jun 20	1.2 d
85C00544	Inactive	Jun 19	Moderate
	Rough Coat	Jun 19	Slight
	Tremors	Jun 19	Slight
	Death	Jun 20	20.4 h
85C00545	Inactive	Jun 19,20	Moderate
	Rough Coat	Jun 19,20	Slight
	Tremors	Jun 19,20	Moderate
	Material, Red, Abdomen	Jun 20	Moderate
	Decr. Temperature	Jun 20	N/A
	Death	Jun 21	1.9 d
85C00548	Inactive	Jun 19	Moderate
	Rough Coat	Jun 19	Slight
	Stain, Yellow, Perianal	Jun 19	Slight
	Tremors	Jun 19	Slight
	Urine, Bloody	Jun 19	Present
	Gasping	Jun 19	Slight
	Death	Jun 20	21.9 h

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: 398 mg/kg Compound 1 (cont.)

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00549	Inactive	Jun 19	Moderate
	Rough Coat	Jun 19	Moderate
	Stain, Yellow, Perianal	Jun 19	Slight
	Stain, Yellow, Abdomen	Jun 19	Moderate
	Urine, Bloody	Jun 19	Present
	Death	Jun 20	21.8 h
85C00554	Inactive	Jun 19,20	Moderate
	Rough Coat	Jun 19	Slight
	Tremors	Jun 20	Slight
	Hypotonia	Jun 20	Slight
	Material, Red, Abdomen	Jun 20	Moderate
	Decr. Temperature	Jun 20	N/A
	Urine, Bloody	Jun 20	Present
	Death	Jun 21	1.8 d
85C00577	Inactive	Jun 19,20	Moderate
	Tremors	Jun 19,20	Slight
	Urine, Bloody	Jun 19,20	Present
	Rough Coat	Jun 19,20	Slight
	Material, Red, Abdomen	Jun 20	Moderate
	Decr. Temperature	Jun 20	N/A
	Death	Jun 21	1.8 d
85C00589	Inactive	Jun 19,20	Moderate
	Tremors	Jun 19,20	Slight
	Urine, Bloody	Jun 19	Present
	Diarrhea	Jun 20	Slight
	Decr. Temperature	Jun 20	N/A
	Death	Jun 21	1.8 d

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: 501 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00552	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Tremors	Jun 17	Slight
	Jumping	Jun 17	Slight
	Penis, Bloody	Jun 17	Present
	Death	Jun 18	19.5 h
85C00557	Inactive	Jun 17	Marked
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Slight
	Death	Jun 18	19.5 h
85C00558	Inactive	Jun 17	Moderate
	Rough Coat	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Penis, Bloody	Jun 17	Present
	Death	Jun 18	19.5 h
85C00559	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Slight
	Death	Jun 18	19.5 h
85C00562	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Slight
	Material, Yellow, Perianal	Jun 17	Slight
	Tremors	Jun 17	Slight
	Penis, Bloody	Jun 17	Present
	Death	Jun 18	19.5 h

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: 501 mg/kg Compound 1 (cont.)

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00570	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Moderate
	Decr. Temperature	Jun 17	N/A
	Tremors	Jun 17	Slight
	Death	Jun 18	19.5 h
85C00571	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
	Rough Coat	Jun 17	Slight
	Stain, Yellow, Abdomen	Jun 17	Slight
	Penis, Bloody	Jun 17	Present
	Death	Jun 18	19.5 h
85C00572	Inactive	Jun 17	Marked
	Rough Coat	Jun 17	Moderate
	Hunched Posture	Jun 17	Moderate
	Depr. Righting Reflex	Jun 17	Slight
	Decr. Temperature	Jun 17	Marked
	Material, Yellow, Abdomen	Jun 17	Moderate
	Tremors	Jun 17	Slight
	Penis, Bloody	Jun 17	Present
	Death	Jun 17	3.9 h
85C00580	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Moderate
	Jumping	Jun 17	Slight
	Death	Jun 18	19.4 h
85C00581	Inactive	Jun 17	Moderate
	Hunched Posture	Jun 17	Slight
	Tremors	Jun 17	Slight
	Penis, Bloody	Jun 17	Present
	Death	Jun 18	19.4 h

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

MALE: Vehicle Control

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00536	Normal	N/A	N/A
85C00555	Normal	N/A	N/A
85C00561	Normal	N/A	N/A
85C00564	Normal	N/A	N/A
85C00578	Normal	N/A	N/A
85C00579	Normal	N/A	N/A
85C00583	Normal	N/A	N/A
85C00584	Rough Coat	Jun 19	Slight
	Material, Yellow, Perianal	Jun 21-23	Slight
85C00588	Hunched Posture	Jun 19	Slight
85C00590	Normal	N/A	N/A

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 159 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00599	Normal	N/A	N/A
85C00613	Hunched Posture	Jun 17	Slight
85C00619	Inactive	Jun 17	Slight
85C00624	Hunched Posture	Jun 17	Slight
85C00633	Hunched Posture	Jun 17	Slight
	Jumping	Jun 17	Slight
85C00634	Hunched Posture	Jun 17	Slight
85C00643	Hunched Posture	Jun 17	Slight
85C00645	Hunched Posture	Jun 17	Slight
	Stain, Yellow, Mouth	Jun 17	Slight
85C00649	Normal	N/A	N/A

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 251 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00596	Hunched Posture	Jun 17	Slight
85C00600	Hunched Posture	Jun 17	Slight
85C00602	Normal	N/A	N/A
85C00605	Hunched Posture	Jun 17	Slight
85C00607	Normal	N/A	N/A
85C00616	Normal	N/A	N/A
85C00617	Normal	N/A	N/A
85C00622	Hunched Posture	Jun 17	Slight
85C00639	Normal	N/A	N/A
85C00650	Inactive Hunched Posture	Jun 17 Jun 17	Slight Slight

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 316 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00595	Inactive	Jun 19	Slight
85C00603	Inactive	Jun 19	Slight
	Hunched Posture	Jun 19,20	Slight
	Death	Jun 21	1.8 d
85C00609	Hunched Posture	Jun 19	Slight
	Urine, Bloody	Jun 19	Present
85C00615	Inactive	Jun 19	Slight
85C00620	Inactive	Jun 19	Marked
	Depr. Grasping Reflex	Jun 19	Marked
	Depr. Righting Reflex	Jun 19	Marked
	Gasping	Jun 19	Slight
	Material, Red, Nose	Jun 19	Moderate
	Death	Jun 19	4.6 h
85C00621	Inactive	Jun 19,20	Slight
	Hunched Posture	Jun 19	Slight
85C00636	Hunched Posture	Jun 19	Slight
85C00647	Hunched Posture	Jun 19,20	Slight
	Urine, Bloody	Jun 19	Present
	Inactive	Jun 20	Slight
85C00652	Inactive	Jun 19-21	Marked
	Tremors	Jun 19-21	Moderate
	Diarrhea	Jun 20	Slight
	Decr. Temperature	Jun 20,21	N/A
	Material, Red, Perianal	Jun 21	Moderate
	Death	Jun 22	2.9 d
85C00654	Inactive	Jun 19,20	Slight
	Tremors	Jun 19,20	Marked
	Decr. Temperature	Jun 19,20	N/A
	Material, Red, Perianal	Jun 20	Marked
	Death	Jun 21	1.9 d

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 398 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00606	Inactive	Jun 17,18	Moderate
	Hunched Posture	Jun 17-20	Moderate
	Death	Jun 21	3.9 d
85C00611	Inactive	Jun 17,18	Moderate
	Hunched Posture	Jun 17,18	Moderate
	Tremors	Jun 17,18	Slight
	Material, Red, Abdomen	Jun 18	Moderate
	Death	Jun 19	1.8 d
85C00618	Hunched Posture	Jun 17,18	Moderate
	Inactive	Jun 17,18	Moderate
85C00625	Inactive	Jun 17-19	Marked
	Hunched Posture	Jun 17,18	Moderate
	Incr. Respiration Rate	Jun 17	Slight
	Material, Red, Perianal	Jun 17,18	Slight
	Tremors	Jun 17-19	Slight
	Decr. Temperature	Jun 18,19	N/A
	Death	Jun 20	2.8 d
85C00630	Inactive	Jun 17,18	Marked
	Hunched Posture	Jun 17	Slight
	Tremors	Jun 17,18	Moderate
	Urine, Red	Jun 18	Present
	Decr. Temperature	Jun 18	N/A
	Death	Jun 19	1.8 d
85C00632	Inactive	Jun 17,18	Marked
	Hunched Posture	Jun 17,18	Moderate
	Tremors	Jun 18	Moderate
	Strain, Red, Moderate	Jun 18	Moderate
	Decr. Temperature	Jun 18	N/A
	Death	Jun 19	1.8 d

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 398 mg/kg Compound 1 (cont.)

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00638	Inactive	Jun 17-19	Marked
	Hunched Posture	Jun 17,18	Slight
	Tremors	Jun 17-19	Moderate
	Decr. Temperature	Jun 17-19	N/A
	Urine, Dark Red	Jun 18,19	Present
	Material, Yellow, Mouth	Jun 19	Moderate
	Death	Jun 20	2.8 d
85C00646	Inactive	Jun 17-19	Marked
	Hunched Posture	Jun 17-19	Slight
	Tremors	Jun 17-19	Moderate
	Decr. Temperature	Jun 17-19	N/A
	Death	Jun 20	2.8 d
85C00648	Inactive	Jun 17	Slight
	Hunched Posture	Jun 17	Slight
	Tremors	Jun 17	Slight
	Death	Jun 18	19.5 h
85C00655	Inactive	Jun 17	Marked
	Hunched Posture	Jun 17	Slight
	Stain, Yellow, Mouth	Jun 17	Moderate
	Tremors	Jun 17	Slight
	Death	Jun 18	19.5 h

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 501 mg/kg Compound 1

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00594	Inactive	Jun 19	Marked
	Tremors	Jun 19	Slight
	Death	Jun 20	20.3 h
85C00598	Inactive	Jun 19	Moderate
	Hunched Posture	Jun 19	Slight
	Tremors	Jun 19	Slight
	Death	Jun 19	4.3 h
85C00608	Death	Jun 19	2.8 h
85C00610	Inactive	Jun 19	Moderate
	Tremors	Jun 19	Slight
	Death	Jun 20	20.9 h
85C00626	Inactive	Jun 19	Slight
	Tremors	Jun 19	Slight
	Death	Jun 19	4.2 h
85C00627	Inactive	Jun 19	Slight
	Hunched Posture	Jun 19	Slight
	Jumping	Jun 19	Slight
	Urine, Bloody	Jun 19	Present
	Feces, Bloody	Jun 19	Present
	Death	Jun 20	20.9 h
85C00635	Death	Jun 19	4.2 h
85C00641	Inactive	Jun 19	Moderate
	Hunched Posture	Jun 19	Slight
	Tremors	Jun 19	Slight
	Prostrate	Jun 19	Present
	Urine, Bloody	Jun 19	Present
	Death	Jun 19	4.2 h

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: 501 mg/kg Compound 1 (cont.)

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00642	Inactive	Jun 19	Marked
	Tremors	Jun 19	Slight
	Lacrimation	Jun 19	Slight
	Decr. Temperature	Jun 19	Present
	Depr. Grasping Reflex	Jun 19	Marked
	Depr. Righting Reflex	Jun 19	Marked
	Hypotonia	Jun 19	Marked
	Urine, Bloody	Jun 19	Present
	Feces, Bloody	Jun 19	Present
	Death	Jun 19	20.2 h
85C00644	Inactive	Jun 19	Moderate
	Hunched Posture	Jun 19	Slight
	Tremors	Jun 19	Slight
	Death	Jun 20	20.2 h

Appendix E (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALE: Vehicle Control

Animal Number	Clinical Signs	Dates Observed (1985)	Severity
85C00601	Normal	N/A	N/A
85C00604	Normal	N/A	N/A
85C00612	Normal	N/A	N/A
85C00614	Normal	N/A	N/A
85C00623	Normal	N/A	N/A
85C00631	Normal	N/A	N/A
85C00637	Normal	N/A	N/A
85C00640	Normal	N/A	N/A
85C00651	Normal	N/A	N/A
85C00653	Normal	N/A	N/A

Appendix F: INDIVIDUAL BODY WEIGHTS†

MALES: 200 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00551	26	29	34	35
85C00553	26	30	32	33
85C00563	28	34	38	38
85C00567	29	35	39	38
85C00573	31	34	35	35
85C00576	23	29	32	34
85C00582	28	33	37	39
85C00587	32	36	38	38
85C00591	33	32	36	36
Mean	28.4	32.4	35.7	36.2
Standard Deviation	3.2	2.6	2.6	2.1
Standard Error	1.1	0.9	0.9	0.7

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

MALES: 251 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00547	32	34	35	34
85C00556	25	30	34	35
85C00565	27	32	31	35
85C00566	26	31	33	34
85C00568	32	37	31	36
85C00569	32	35	36	38
85C00575	34	38	33	37
85C00585	25	32	35	35
85C00592	30	33	33	34
85C00593	31	32	35	35
Mean	29.4	33.4	33.6	35.3
Standard Deviation	3.3	2.6	1.7	1.3
Standard Error	1.1	0.8	0.5	0.4

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

MALES: 316 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00534	29	33	Dead	
85C00535	29	32	35	36
85C00537	20	30	34	37
85C00540	29	31	Dead	
85C00542	34	35	Dead	
85C00543	30	35	Dead	
85C00550	30	34	37	39
85C00560	28	31	31	35
85C00574	29	32	Dead	
85C00586	34	38	Dead	
Mean	29.2	33.1	34.2	36.7
Standard Deviation	3.9	2.4	2.5	1.7
Standard Error	1.2	0.8	1.3	0.9

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

MALES: 398 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00533	32	36	Dead	
85C00539	31	37	Dead	
85C00541	26	30	Dead	
85C00544	28	32	Dead	
85C00545	26	33	Dead	
85C00548	27	34	Dead	
85C00549	31	34	Dead	
85C00554	29	31	Dead	
85C00577	28	33	Dead	
85C00589	35	41	Dead	
Mean	29.3	34.1		
Standard Deviation	2.9	3.2		
Standard Error	0.9	1.0		

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

MALES: 501 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00552	28	30	Dead	
85C00557	29	34	Dead	
85C00558	32	35	Dead	
85C00559	28	33	Dead	
85C00562	28	31	Dead	
85C00570	28	33	Dead	
85C00571	26	33	Dead	
85C00572	31	35	Dead	
85C00580	36	39	Dead	
85C00581	27	31	Dead	
Mean	29.3	33.4		
Standard Deviation	3.0	2.6		
Standard Error	0.9	0.8		

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

MALES: Vehicle Control

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00536	26	31	37	37
85C00555	32	34	37	37
85C00562	27	30	32	34
85C00564	32	33	36	34
85C00578	31	33	35	35
85C00579	29	35	40	38
85C00583	24	29	32	33
85C00584	33	35	40	38
85C00588	30	33	36	36
85C00590	31	34	38	38
Mean	29.4	32.7	36.3	36.0
Standard Deviation	3.0	2.1	2.8	1.9
Standard Error	0.9	0.7	0.9	0.6

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

FEMALES: 159 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00599	29	29	30	33
85C00613	28	30	31	32
85C00619	24	27	30	30
85C00624	27	27	29	29
85C00633	29	29	30	29
85C00634	34	35	34	35
85C00643	26	28	30	29
85C00645	24	29	29	30
85C00649	29	29	32	31
Mean	27.8	29.2	30.6	30.9
Standard Deviation	3.1	2.4	1.6	2.1
Standard Error	1.0	0.8	0.5	0.7

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

FEMALES: 251 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00596	24	29	31	30
85C00600	24	27	29	28
85C00602	27	30	30	31
85C00605	27	28	31	31
85C00607	25	28	28	28
85C00616	24	26	28	29
85C00617	27	30	32	33
85C00622	25	24	24	24
85C00639	28	28	30	30
85C00650	34	36	31	33
Mean	26.5	28.6	29.4	29.7
Standard Deviation	3.0	3.2	2.3	2.7
Standard Error	1.0	1.0	0.7	0.8

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

FEMALES: 316 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00595	23	29	29	27
85C00603	28	28	Dead	
85C00609	23	29	31	31
85C00615	24	26	27	26
85C00620	25	26	Dead	
85C00621	27	28	29	30
85C00636	27	26	30	27
85C00647	24	26	27	26
85C00652	24	27	Dead	
85C00654	27	29	Dead	
Mean	25.2	27.4	28.8	27.8
Standard Deviation	1.9	1.4	1.6	2.1
Standard Error	0.6	0.4	0.7	0.9

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

FEMALES: 398 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00606	28	28	Dead	
85C00611	27	27	Dead	
85C00618	24	25	24	27
85C00625	28	28	Dead	
85C00630	25	29	Dead	
85C00632	31	33	Dead	
85C00638	30	31	Dead	
85C00646	27	31	Dead	
85C00648	27	27	Dead	
85C00655	30	32	Dead	
Mean	27.7	29.1	24.0	27.0
Standard Deviation	2.2	2.6		
Standard Error	0.7	0.8		

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

FEMALES: 501 mg/kg Compound 1

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00594	26	27	Dead	
85C00598	24	31	Dead	
85C00608	29	32	Dead	
85C00610	27	28	Dead	
85C00626	28	29	Dead	
85C00627	25	28	Dead	
85C00635	29	29	Dead	
85C00641	26	24	Dead	
85C00642	28	28	Dead	
85C00644	26	29	Dead	
Mean	26.8	28.5		
Standard Deviation	1.7	2.2		
Standard Error	0.5	0.7		

† Weight is given in grams.

Appendix F (cont.): INDIVIDUAL BODY WEIGHTS†

FEMALES: Vehicle Control

Animal Number	Receipt	Dosing	Day 7	Day 14
85C00601	29	30	30	31
85C00604	34	33	37	36
85C00612	29	31	33	32
85C00614	25	25	27	27
85C00623	31	32	32	34
85C00631	27	31	32	32
85C00637	26	27	29	28
85C00640	27	28	28	28
85C00651	28	31	33	32
85C00653	26	28	28	30
Mean	28.2	29.6	30.1	31.0
Standard Deviation	2.7	2.5	3.1	2.8
Standard Error	0.9	0.8	1.0	0.9

† Weight is given in grams.

Appendix G: PATHOLOGY REPORT

Pathology Report
Acute Oral Toxicity Study (LD_{50})
GLP Study 85028

Test substance: Water Disinfectant Compound I.

Species: Mouse. Strain: ICR.

Investigator: Dr. Gerald Hiatt, Toxicology Branch

History: Refer to LAIR SOP-OP-STX-36. Animals that did not die were killed with sodium pentobarbital anesthesia and axillary bleeding.

Microscopic findings:

PATH ACC#	ANIMAL ID#	MORPHOLOGIC DX
37908	85C00533	Lung, spinal cord, adipose tissue - Not remarkable (NR)
37924	85C00599	Pancreas - NR
37915	85C00560	Nephritis, interstitial, chronic, multifocal, moderate, kidney
37953	85C00568	Hyperplasia, lymphoid, myeloid, spleen. Hepatitis & pancreatitis with abscessation, liver, pancreas. Peritonitis & fibrosis, focal, stomach.
37955	85C00575	Hyperplasia, lymphoid, myeloid, spleen. Hepatitis with abscessation, liver.
37959	85C00584	Cystic duct, preputial gland
37932	85C00618	Hepatitis with abscessation with plant material and hair. Peritonitis, chronic, stomach.

Appendix G (cont.): PATHOLOGY REPORT

Pathology Report
GLP Study 85028

Gross findings:

PATH ACC#	ANIMAL ID#	GROSS FINDINGS
	GROUP 1M/MALE/200 mg/kg	
37913	85C00551	Live - NR
37914	85C00553	Live - NR
37916	85C00561	Live - NR
37917	85C00567	Live - NR
37918	85C00573	Live - NR
37919	85C00576	Live - NR
37920	85C00582	Live - NR
37921	85C00587	Live - NR
37922	85C00591	Live - NR
	GROUP 2M/MALE/316 mg/kg	
37876	85C00534	Dead - NR
37910	85C00535	Live - NR
37911	85C00537	Live - NR
37856	85C00540	Dead - NR
37857	85C00542	Dead - NR
37858	85C00543	Dead - NR
37912	85C00550	Live - NR
37915	85C00560	Live: Kidneys - granular cortical surfaces
37887	85C00574	Dead - NR
37872	85C00566	Dead - NR
	GROUP 3M/MALE/501 mg/kg	
37859	85C00552	Dead - NR
37860	85C00557	Dead - NR
37861	85C00558	Dead - NR
37862	85C00559	Dead - NR
37863	85C00562	Dead - NR
37864	85C00570	Dead - NR
37865	85C00571	Dead - NR
37866	85C00572	Dead - NR
37867	85C00580	Dead - NR
37868	85C00581	Dead - NR

Appendix G (cont.): PATHOLOGY REPORT

Pathology Report
GLP Study 85028

GROUP 4M/MALE/251 mg/kg

PATH ACC#	ANIMAL ID#	GROSS FINDINGS
37946	85C00547	Live - NR
37948	85C00556	Live - NR
37951	85C00565	Live - NR
37952	85C00566	Live - NR
37953	85C00568	Live - Adhesions between liver, stomach, ileum splenomegally
37954	85C00569	Live - NR
37955	85C00575	Live: Liver - multiple abscesses, splenomegally
37960	85C00585	Live - NR
37963	85C00592	Live - NR
37964	85C00593	Live - NR

GROUP 5M/MALE/398 mg/kg

37908	85C00533	Dead: Scapula - overlying area red.
37897	85C00539	Dead - NR
37898	85C00541	Dead - NR
37885	85C00544	Dead - NR
37899	85C00545	Dead - NR
37896	85C00548	Dead - NR
37886	85C00549	Dead - NR
37900	85C00554	Dead - NR
37901	85C00577	Dead - NR
37902	85C00589	Dead - NR

GROUP 6M/MALE/Vehicle

37945	85C00536	Live - NR
37947	85C00555	Live - NR
37949	85C00561	Live - NR
37950	85C00564	Live - NR
37956	85C00578	Live - NR
37957	85C00579	Live - NR
37958	85C00583	Live - NR
37959	85C00584	Live - Preputial gland mass (cyst)
37961	85C00588	Live - NR
37962	85C00590	Live - NR

Appendix G (cont.): PATHOLOGY REPORT

Pathology Report
GLP Study 85028

GROUP 1F/FEMALE/159 mg/kg

PATH ACC#	ANIMAL ID#	GROSS FINDINGS
37924	85CB0599	Live: Pancreas - edematous & red (probable pentobarbital injection site)
37929	85CB0613	Live - NR
37933	85CB0619	Live - NR
37935	85CB0624	Live - NR
37936	85CB0633	Live - NR
37937	85CB0634	Live: Ovarian cyst, left(4mm diameter)
37939	85CB0643	Live - NR
37940	85CB0645	Live - NR
37941	85CB0649	Live - NR

GROUP 2F/FEMALE/251 mg/kg

37923	85CB0596	Live - NR
37925	85CB0609	Live - NR
37926	85CB0612	Live - NR
37927	85CB0615	Live - NR
37928	85CB0617	Live - NR
37930	85CB0616	Live - NR
37931	85CB0617	Live - NR
37934	85CB0622	Live - NR
37938	85CB0639	Live - NR
37942	85CB0650	Live - Adhesions between liver & stomach

GROUP 3F/FEMALE/398 mg/kg

37904	85CB0606	Dead - NR
37873	85CB0611	Dead - NR
37932	85CB0618	Live - Adhesions between liver & stomach
37890	85CB0625	Dead - NR
37874	85CB0630	Dead - NR
37875	85CB0632	Dead - NR
37892	85CB0638	Dead - NR
37895	85CB0646	Dead - NR
37869	85CB0648	Dead - NR
37870	85CB0655	Dead - NR

Appendix G (cont.): PATHOLOGY REPORT

Pathology Report
GLP Study 85028

GROUP 4F/FEMALE/316 mg/kg

PATH ACC#	ANIMAL ID#	GROSS FINDINGS
37965	85C00595	Live - NR
37903	85C00603	Dead - NR
37968	85C00609	Live - NR
37971	85C00615	Live - NR
37880	85C00620	Dead - NR
37972	85C00621	Live - NR
37975	85C00636	Live - NR
37978	85C00647	Live - NR
37906	85C00652	Dead - NR
37905	85C00654	Dead - NR

GROUP 5F/FEMALE/501 mg/kg

37888	85C00594	Dead - NR
37878	85C00598	Dead - NR
37879	85C00608	Dead - NR
37889	85C00610	Dead - NR
37881	85C00626	Dead - NR
37891	85C00627	Dead - NR
37882	85C00635	Dead - NR
37883	85C00641	Dead - NR
37893	85C00642	Dead - NR
37894	85C00644	Dead - NR

GROUP 6F/FEMALE/Vehicle

37966	85C00601	Live - NR
37967	85C00604	Live - NR
37969	85C00612	Live - NR
37970	85C00614	Live - NR
37973	85C00623	Live - NR
37974	85C00631	Live - NR
37967	85C00637	Live - NR
37977	85C00640	Live - NR
37979	85C00651	Live - NR
37980	85C00653	Live - NR

Appendix G (cont.): PATHOLOGY REPORT

Pathology Report
GLP Study 85028

SUMMARY TABLE - MALES

GROUP	DOSE (mg/kg)	NO.	DEATHS	%MORTALITY	TIME TO DEATH <1 DAY	TIME TO DEATH 2-6 DAYS
6M	0	10	0	0	0	0
1M	200	9	0	0	0	0
4M	251	10	0	0	0	0
2M	316	10	6	60	3	3
5M	398	10	10	100	5	5
3M	501	10	10	100	10	0

SUMMARY TABLE - FEMALES

GROUP	DOSE (mg/kg)	NO.	DEATHS	%MORTALITY	TIME TO DEATH <1 DAY	TIME TO DEATH 2-4 DAYS
6F	0	10	0	0	0	0
1F	159	9	0	0	0	0
2F	251	10	0	0	0	0
4F	316	10	4	40	1	3
3F	398	10	9	90	2	7
5F	501	10	10	100	10	0

Gross Comments:

1. Dose related toxicity of the compound is apparent.

2. All gross and microscopic lesions were considered incidental findings, not related to the test compound. The abscesses noted in livers and pancreas and the peritonitis localized around the stomach in four animals were most likely due to iatrogenic laceration of the stomach wall with the dosing syringe. The splenomegally was a reactive response to the chronic intra-abdominal inflammation. Red tissues overlying the scapula of mouse 85D00533 were probably induced while being held for dosing and are traumatic in origin.

3. Clostridia perfringens, Pasteurella multocida, Escherichia coli, and Enterococci sp. were cultured and identified from the abdominal abscesses of two mice (85D00568 and 85D00575).

G. TRACY MINOTC, DVM
MAJ, VC
Comparative Pathology Branch
31 January 1986

OFFICIAL DISTRIBUTION LIST

Commander
US Army Medical Research
& Development Command
ATTN: SGRD-RMS/Mrs. Madigan
Fort Detrick, MD 21701-5012

Defense Technical Information Center
ATTN: DTIC/DDAB (2 copies)
Cameron Station
Alexandria, VA 22304-6145

Office of Under Secretary of Defense
Research and Engineering
ATTN: R&AT (E&LS), Room 3D129
The Pentagon
Washington, DC 20301-3080

DASG-AAFJML
Army/Air Force Joint Medical Library
Offices of the Surgeons General
5109 Leesburg Pike, Room 670
Falls Church, VA 22041-3258

HQ DA (DASG-ZXA)
WASH DC 20310-2300

Commandant
Academy of Health Sciences
US Army
ATTN: HSHA-CDM
Fort Sam Houston, TX 78234-6100

Uniformed Services University of
Health Sciences
Office of Grants Management
4301 Jones Bridge Road
Bethesda, MD 20814-4799

US Army Research Office
ATTN: Chemical and Biological
Sciences Division
PO Box 12211
Research Triangle Park, NC 27709-2211

Director
Walter Reed Army Institute of Research
ATTN: SGRD-UWZ-L
Washington, DC. 20307-5100

Commander
US Army Medical Research Institute
of Infectious Diseases
ATTN: SGRD-ULZ-A
Fort Detrick, MD 21701-5011

Commander
US Army Medical Materiel Development
Activity
Fort Detrick, Bldg T622
Frederick, MD 21701-5009

Commander
US Army Biomedical Research and
Development Laboratory
ATTN: Library
Fort Detrick, Bldg 568
Frederick, MD 21701-5010

Commander
US Army Research Institute of
Environmental Medicine
ATTN: SGRD-UE-RSA
Kansas Street
Natick, MA 01760-5007

Commander
US Army Institute of Surgical Research
Fort Sam Houston, TX 78234-6200

Commander
US Army Medical Research Institute of
Chemical Defense
ATTN: SGRD-UV-AJ
Aberdeen Proving Ground, MD 21010-5425

Commander
US Army Aeromedical Research
Laboratory
Fort Rucker, AL 36362-5000

AIR FORCE Office of Scientific
Research (NL)
Building 410, Room A217
Bolling Air Force Base, DC 20332-6448

USAF School of Aerospace Medicine
Document Section
USAFSAM/TSKD
Brooks Air Force Base, TX 78235-5301

Head, Biological Sciences Division
OFFICE OF NAVAL RESEARCH
800 North Quincy Street
Arlington, VA 22217-5000